The adaptation of muscle activity during split-belt walking reveals age-dependent decline of motor learning.

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Abstract

We are constantly adjusting our movements to changes in our body or environment through online feedback or planned feedforward processes. It is thought that only feedforward motor commands are adapted in locomotion and underlie the observed aftereffects upon experiencing a novel situation, such as split-belt walking with the legs moving at different speeds. Feedback (a.k.a., reactive) commands are assumed to simply reflect the walking condition by suddenly changing when the novel environment is introduced or removed. However, this has been concluded based on kinematics (strongly influenced by environmental dynamics) and opposes others' suggestion that feedback processes are adapted according to the environment at hand. Thus, we ask if feedback commands are adapted and if this adaptation is dissociated from the recalibration of feedforward ones through the analysis of muscle activity. To this end, we characterized changes in the activity of 15 muscles during and after split-belt walking with respect to regular walking. Importantly, we exploited the temporal dynamics of feedforward and feedback processes to isolate feedback-generated motor commands and characterized its adaptation after short and long exposures to the split condition requiring distinct recalibration of feedforward commands. We found that feedback-mediated motor patterns were adapted. Specifically, subjects exhibited on each leg distinct feedback-generated responses upon introduction of the split perturbation that were mirrored between the legs (i.e., muscle activity of the right was observed on the left and vice versa) when the split condition was removed. This mirroring was only observed after long exposure of the split condition and contrasted the purely ipsilateral changes predicted from environment-dependent feedback activity. Both of which indicated that the mirroring of feedback responses reflected a learning-dependent process. Further, older individuals exhibited limited adaptation of feedback activity, but had equally adapted patterns at the end of the split-belt walking compared to younger participants, suggesting that healthy aging only affects the adaptation of feedback commands. Taken together our results indicate that corrective actions in walking are adapted and that this adaptation is partially dissociated from planned actions, raising the possibility of distinct neural processes underlying the adaptation of feedback- and feedforward-mediated motor control.

Significance statement

We characterized the temporal evolution of muscle activity during a motor adaptation and deadaptation task. We found that feedback-generated motor commands, which react to sudden transitions in the environment, are adapted through prolonged exposure to new environmental conditions. Adapted responses are consistent with subjects adopting the new environment as normal, and the extent of this adaptation is modulated by age. This shows that aftereffects are at least partially a response to the sudden transition in the environment, and not solely the continuation of a motor output optimized for different environmental conditions. Characterizing how muscle activity changes throughout this task and how this predicts motor learning (aftereffects) can aid in the design of individualized protocols to maximize the therapeutic effects of split-belt walking.

1 Introduction

Humans adapt and learn new movements through interactions with the world, but there have been limited efforts investigating the plasticity of muscle activity underlying this motor adaptation. For example, there are several studies in locomotion characterizing changes in kinematics (Reisman et al., 2005, e.g.), kinetics (Ogawa et al., 2014; Sombric et al., 2018), metabolic cost (Finley et al., 2013; Sánchez et al., 2017), and the perception of movements (Vazquez et al., 2015) that are retained following walking on a split-belt treadmill that moves people's legs at different speeds. However, none of the work recording muscle activity provides a detailed characterization of what is adapted in muscle space during and after the novel split-belt situation (Ogawa et al., 2014; Raja et al., 2013; Maclellan et al., 2014). It is important to study the adaptation of muscle activity upon sensorimotor learning because the higher dimension of muscles compared to joints allows distinct motor patterns to produce the same movement (Bernstein, 1967). Thus, changes in muscle activations can reveal the adaptation of richer neural repertoires (Albert and Shadmehr, 2016) that are obscured by purely assessing changes in kinematic or kinetic variables. Therefore, analyzing the adaptation of muscle activity offers the possibility to identify potentially distinct neural control mechanisms through which motor adaptation occurs.

Muscle activity can be adapted through adjusted feedforward- and/or feedback-generated motor commands that contribute distinctively to motor adaptation. Feedforward-generated motor commands are planned ahead of the movement and hence are predictive in nature. Conversely, feedback-generated motor commands arise during movement execution through a transformation of current sensory information into corrective actions (Jordan and Rumelhart, 1992; Bhushan and Shadmehr, 1999). Walking studies presume that only feedforward processes are adapted and underlie the observed aftereffects (Morton and Bastian, 2006; Ogawa et al., 2014). Feedback (a.k.a., reactive) commands are assumed to remain unchanged during locomotor adaptation and simply reflect the walking condition by suddenly changing when the novel environment is introduced or removed (Reisman et al., 2005; Morton and Bastian, 2006; Yokoyama et al., 2018). However, it is unclear whether feedback-generated activity is actually adapted, whether this process is independent of feedforward adaptation, and what impact it has on aftereffects. Dissociating the adaptation (or lack of thereof) feedback and feedforward processes is of

interest as they may reflect distinct learning mechanisms (Wagner and Smith, 2008; Yousif and Diedrichsen, 2012).

It is particularly relevant to evaluate the adaptation of feedback and feedforward commands in middle aged and older individuals because of its implications for movement rehabilitation. Namely, locomotor learning induced by split-belt walking can be exploited to improve the gait of stroke survivors (Reisman et al., 2013; Lewek et al., 2017), whose age mostly ranges between 45 and 80+ years old (Mozaffarian et al., 2016). While there are studies reporting age-related decline in subjects' ability to adapt movements and the retention of those changes (Wolpe et al., 2016; Sombric et al., 2017), it is unknown if healthy aging has a differential impact on feedback and feedforward processes contributing to motor adaptation. Thus, we studied how healthy aging affects the plasticity of muscle activity to gain insights on the learning mechanisms available to patients because of their age.

In sum, we characterized the adaptation of muscle recordings from a broad set of lower-limb muscles during and after split-belt walking. We used sudden changes in muscle activity upon transitions in the walking environment (e.g., split-belts vs. tied-belts) to dissociate the contributions of feedforward and feedback processes to motor adaptation. We found distinct feedback muscle activity following the same environment transition at different adapted states of feedforward-generated motor commands, suggesting that feedback processes are also influenced by the re-calibration of internal representations of walking. Importantly, we were able to predict the adaptation of feedback-generated activity based on each individual's initial muscle activity during the adaptation period. Interestingly, we observed that older subjects adapted less, revealing age-related limitations in sensorimotor learning.

2 Materials and Methods

2.1 Subjects

A group of 15 healthy subjects of ages ranging between 46 to 78 years old (61+-9.9 y.o., 10 female) participated in the study. Their movements and muscle activity were recorded while they experienced a split-belt treadmill adaptation protocol approved by the University of Pittsburgh's Institutional Review Board, and in accordance to the declaration of Helsinski. All subjects provided written informed consent prior to participating in the study.

2.2 Experimental Design

We assessed the adaptation and de-adaptation of muscle activity through the protocol illustrated in Figure 1A. The protocol consisted of treadmill walking in six different conditions presented in the following order: Slow Walking (50 strides), Mid Walking (50 strides), Short Exposure (10 strides), Baseline Mid Walking (150 strides), Adaptation (900 strides, in three blocks of 300) and Post-Adaptation (600 strides, in two blocks of 300 or a single 600 block). Slow and mid walking were used to quantify speed-dependent modulation of muscle activity during regular treadmill walking. Short Exposure and Adaptation were used to differentiate learning-dependent changes in muscle activity that required multiple steps in the split environment from those solely due to unexpected introduction or removal of the split perturbation. Baseline walking was used as a reference for steady state walking on the treadmill prior to motor adaptation. Finally, the Post-Adaptation condition was used to evaluate adaptation aftereffects in muscle activity following split-belt walking. The Adaptation and Post-Adaptation conditions were designed to have enough strides to examine the evolution of muscle activity from a transient to a steady state when the perturbation was either introduced or removed, respectively. These conditions were divided into blocks to minimize fatigue and subjects were instructed not to step between blocks to prevent deadaptation due to unrecorded steps.

Belts moved at different speeds (i.e., 'split' mode) only in the Short Exposure and Adaptation conditions with the dominant leg (self-reported leg used to kick a ball) walking faster. We refer to the dominant leg as the fast leg throughout the text and the non-dominant one as the slow leg. In all other conditions, belts moved at the

same speed relative to one another (i.e., 'tied' mode) (Figure 1A). The treadmill was started and stopped at the beginning and at the end of each condition and speeds did not change while each condition was ongoing. Belts moved at one of three possible speeds throughout the experiment: a self-selected ('mid') walking speed, a 'slow' speed of 66.6% of the mid speed, and a 'fast' speed of 133.3% of the mid speed. We used self-selected speeds to ensure subjects from all ages could complete the entire protocol. The self-selected speed was obtained by first averaging each subject's speed when walking over ground in a 50-meter hallway during 6 minutes (i.e., 6-min walking test (Rikli and Jones, 1998)) and then subtracting $0.35m/s$, which resulted in a comfortable walking speed on a treadmill based on pilot data in older adults ($>$ 65 yrs). Mid speed was $0.72 \pm 0.26m/s$ (mean $\pm$ standard deviation) across the population. Speed values used for all subjects during baseline walking are shown in Table 2.

Safety measures were designed such that participants from older ages could complete the study. First, all subjects wore a harness that only provided weight support in the event of falling but not during walking or standing. Also, subjects were told a few seconds in advance that they could hold on to a handrail (directly located in front of them) whenever a condition or block started or finished. Subjects were encouraged to let go of the handrail as soon as they felt comfortable doing so to minimize the effect of external support on muscle recordings. Finally, we monitored subjects' heart-rate continuously and blood-pressure during the rest breaks to prevent over exertion in any of the participants.

2.3 Data acquisition & analysis

Data collection

We collected electromyographic (EMG) signals, kinematics, and kinetic data to characterize subjects behavior. Surface EMG signals from 15 muscles on each leg were recorded for all subjects (see Table 1 for full list and abbreviations) at 2000Hz using a Delsys Trigno System (Delsys Inc., Natick, Massachussets). Signals were highpass-filtered to remove undesired movement artifacts and then rectified. We used a 2nd order Butterworth filter (dual-pass) with a cutoff frequency of 30Hz, which resulted in 80 dB/dec attenuation and zero-lag (Merletti

and Parker, 2005). Kinematic data was collected at 100 Hz with a passive motion analysis system (Vicon Motion Systems, Oxford, UK). Movements were recorded by placing reflective markers bilaterally on bony landmarks at the ankle (i.e., lateral malleolus) and the hip (i.e., greater trochanter). Ground reaction forces were recorded with an instrumented split-belt treadmill (Bertec Corporation, Columbus, Ohio) and sampled at 1000Hz. Forces along the axis of gravity (Fz) were used to determine when the foot landed (i.e., heel-strike: $Fz > 10N$) or was lifted off the ground (i.e., toe-off: $Fz < 10N$).

Data Analysis

EMG parameters: We characterized the changes in EMG during and after split-belt walking by first generating muscle activity variables that discretized each muscle's activity over the gait cycle (Figure 1B). Filtered EMG activity was divided in sub-intervals of the gait cycle aligned to gait events to focus on changes in muscle activity within the gait cycle, rather than on activation changes due to differences in timing of the gait cycle across the distinct walking conditions (Dietz et al., 1994; Reisman et al., 2005). For example, in Figure 1B we present sample EMG traces for baseline walking at medium speed (gray trace) and late adaptation (green traces) for the leg walking slow (top panel) and the one walking fast (bottom panel). More specifically, we divided the gait cycle of each leg into 6 intervals according to well defined gait phases (Perry and Burnfield, 2010): first double-support (from ipsilateral heel-strike to contralateral toe-off), early single-stance (first half of single stance), late single-stance (second half of single stance), second double support (from contralateral heel-strike to ipsilateral toe-off), early swing (first half of swing phase) and late swing (second half of swing phase). In order to achieve better temporal resolution, each of these 6 intervals where further divided into two equal sub-intervals, yielding 12 intervals for each gait cycle. Note that the normalized gait cycles appear to be the same duration because of our normalization procedure. However, each of these sub-intervals' duration ranged between 75 and 120 ms. The precise timing for each sub-interval throughout the different epochs of the study are presented in Table 3. Muscle activity amplitude was averaged in time for each of these sub-intervals, for every stride and muscle resulting in 360 muscle activity variables per subject: 12 intervals x 30 muscles. Sample of these variables for one muscle in one gait cycle for Baseline (B) (gray row) and late Adaptation (lA)

(green row) are presented in the rows below the EMG trances in Figure 1B.

These EMG variables were normalized using the activity during Baseline walking as a reference. To this end, we first computed the mean activity for each sub-interval across the last 40 strides (i.e., steady state) of Baseline walking. Then each muscle's activity was scaled such that the least and most active phases for every muscle during baseline had the value of 0 and 1, respectively. We specifically subtracted the mean value for the least active phase in Baseline from the activity in all sub-intervals for all strides in all conditions and we subsequently divided the shifted values by the mean baseline value for the most active phase. Following this normalization, the units of EMG activity for each muscle of a given subject were represented with respect to the average maximum value recorded in that muscle during Baseline walking of the same subject. This scaling allowed us to aggregate subjects and compare effect sizes across muscles even when recorded EMG amplitudes were very different because of sensor placement or underlying tissue properties (Figure 1D).

Kinematic parameters: The adaptation of movements was characterized with step-length asymmetry, which is a metric known to change during and after split-belt walking (Reisman et al., 2005). We computed step-length asymmetry on each stride cycle by calculating the difference in step lengths (i.e., ankle to ankle distance at foot landing) for two consecutive steps taken with the fast and slow leg. This difference was normalized by the sum of step lengths to obtain a measure that was a proportion of each subjects' step sizes. A zero step-length asymmetry value indicated that steps lengths were even, negative values indicated that the (non-dominant) leg walking on the slow belt was taking longer steps than the (dominant) one on the fast belt and viceversa for positive values.

We also computed body displacement with respect to the foot in contact with the ground during the stance phase for each leg. This was done to interpret the changes in muscle activity upon transitions between tied and split conditions. Body displacement was computed as the anterior-posterior distance between the middle of the hip markers (greater trochanter) and the ankle from ipsilateral heel-strike to contra-lateral heel-strike. To focus on displacement changes within this period of the stance phase, we aligned and divided the body displacement in two sub-intervals: double support (from ipsilateral heel-strike to contra-lateral toe off) and single stance (from

contra-lateral toe-off to contra-lateral heel-strike). This normalization of the time axis facilitated the comparison of body displacement during stance of the fast vs. the slow leg or across walking conditions given the distinct stance durations between legs and treadmill modes (tied vs. split) (Reisman et al., 2005).

Outcome measures: EMG and kinematic parameters were used to characterize the behavior during six experimental epochs (Figure 1): Short Exposure (all strides except first and last), early and late Baseline, early and late Adaptation, and early Post-Adaptation. 'Early' epochs were characterized by the average of the initial 15 strides and 'late' epochs by the average of the last 40 strides for each condition. Recordings during the very first and last stride of each condition were excluded to eliminate from the analysis effects linked to the treadmill's start and stop. For example, early Adaptation consisted of the mean activity for strides 2-16, such that 15 strides were considered but the first one was excluded.

We compared muscle activity for a given epoch with that of Baseline walking (Figure 1D). This allowed us to characterize how motor commands differed from normal walking along the adaptation process. The comparison was done by calculating differences in muscle activation variables between an epoch of interest and those during late Baseline walking (e.g., $lA_B = lA - B$ in Figure 1B). We also computed differences in muscle activation variables between consecutive epochs in which subjects transitioned from the split-to-tied (e.g., $eP_{lA} = eP - lA$) or tied-to-split conditions (e.g., $eA_B = eA - B$). The magnitude of these was calculated by computing the euclidean norm of the 360-dimensional vectors (e.g., $\|lA_B\|$). This offered a quantification of overall changes in muscle activity with one single metric, which was particularly relevant for eP_B since it indicated the aftereffect size in muscle space.

2.4 Characterizing feedback components of muscle activity

Previous studies have dissociated the adaptation of feedforward and feedback commands through restriction of limb trajectories when error signals are known (Scheidt et al., 2000; Wagner and Smith, 2008; Albert and Shadmehr, 2016), which is not the case for walking. Thus, we propose to dissociate the adaptation of feedforward and feedback components of muscle activity through an operational definition that exploits the

distinct temporal dynamics of these processes. We specifically assume that feedforward components change slowly upon experiencing a novel situation because they are generated purely through internal models that are updated gradually. On the other hand, feedback components can change rapidly after a transition in the walking environment since sensory information is available immediately following a change in the walking context. Note that our definition of feedback-generated motor commands includes modulation of muscle activity that occurs at different latencies after a movement disturbance (i.e., short- and long-latency reflex responses and subsequent voluntary responses) (Horak et al., 1990). In sum, we quantified feedback-generated activity as the changes in EMG after a sudden environmental transition (i.e., EMG_{after}) with respect to EMG activity before the transition (i.e., EMG_{before}). In equation form:

$$\Delta EMG = EMG_{after} - EMG_{before} = \text{FEEDBACK-GENERATED ACTIVITY}$$

Thus, even if the change in EMG is strictly equal to the change in both feedback and feedforward-generated activity, we approximated the latter one to be negligible because it occurs at a slower rate compared to changes in feedback-generated activity after an abrupt transition in the walking condition. As an example, in the split-to-tied transition this results in:

$$\Delta EMG_{split-to-tied} = eP_B - lA_B = eP_{lA} \tag{1}$$

Where lA and eP are the muscle activity before and after the split-to-tied transition, respectively. This equation simply formalizes the idea that feedback-generated activity might change immediately after removing the split-belts environment. Importantly, the muscle activity aftereffects ($eP_B = eP - B$) are not the same as our definition of the feedback-generated activity upon removal of the split condition (Eq. 1), but contribute to it. Namely, we anticipate that aftereffects will be composed of the learned motor pattern carried over from late Adaptation to Postadaptation (i.e., feedforward component) plus a feedback component due to the sudden environmental transition.

We analyzed the structure of the feedback component of aftereffects (eP_{lA}) to gain insight into learning-dependent mechanisms. More specifically, we considered three possibilities for the feedback component of aftereffects in muscle space (Figure 1C): 1) negligible relative to feedforward commands (H1), 2) solely environment-dependent

(non-adaptive) feedback responses (H2), and 3) adaptive feedback responses (H3). The basis and rationale for each one of our hypothesis is presented in the following sections.

2.4.1 Hypothesis 1 (H1): Negligible feedback compared to feedforward contributions to after-effects

One possibility is that muscle activity is mostly controlled in a feedforward manner when the split perturbation is removed. By mostly, we mean that feedback responses are negligible in magnitude when compared to the feedforward components. In other words, muscle activity before (lA_B) and after (eP_B) the split-to-tied transition is roughly the same, as illustrated in the schematic of H1 in Figure 1C. This hypothesis suggests that feedback components are present during the tied-to-split transition (eA_B), but are greatly reduced during the split-to-tied transition because the return to a familiar environment (i.e., tied condition) may not require the same extent of feedback responses as when a novel condition (i.e., split) is experienced. We formulated this hypothesis on the basis that aftereffects result from the continuation of the motor output updated during the Adaptation period (Malone et al., 2012). Formally expressed:

$$\Delta EMG_{split-to-tied} = eP_B - lA_B = 0 \;\Rightarrow\; eP_B = lA_B$$

2.4.2 Hypothesis 2 (H2): Unadaptive feedback-generated motor commands

Another possibility is that feedback responses are not negligible during post-adaptation, but they are not adaptive in nature. That is, the feedback processes generating them are not updated during split-belt walking and produce corrective muscle patterns solely as a function of the sensed environment. This hypothesis makes two predictions. First, feedback-generated activity would change similarly after removing the split perturbation following a long or short exposure to the split condition because the environment transitions identically in these two situations. Second, we would expect that changes in muscle activity originated by the tied-to-split transition (i.e., eA_B) would be numerically opposite from those from the split-to-tied transition (Figure 1C, H2 schematic) because the environment changes in exactly the opposite manner in these two transitions. Thus, any muscle whose activity increased when the split perturbation was introduced, should decrease by the same amount when it is removed (and vice versa). We reasoned that this hypothesis would be plausible given that

step length asymmetry (conventionally used to characterize locomotor adaptation and after-effects) is perturbed during early adaptation and early post-adaptation by approximately the same amount but in opposite directions (Reisman et al., 2005). Formally expressed:

$$\Delta EMG_{split-to-tied} = eP_B - lA_B = -eA_B \;\; \Rightarrow \;\; eP_B = lA_B - eA_B$$

Where eA_B represents muscle activity during the Early Adaptation with respect to Baseline. These aftereffects (eP_B) are exactly what would be predicted from a linear model whose input is environmental condition and its output is muscle activity.

2.4.3 Hypothesis 3 (H3): Feedback responses adapt to a "new normal"

Finally, we considered the possibility that substantial feedback responses are not only present following transitions in walking speeds, but they are adaptive. According to this hypothesis, changes in activity from the split-to-tied conditions would be different after a short vs. a long exposure to the split environment. We specifically considered that following the long exposure, the motor system would learn that the split (perturbed) condition is the "new normal" and hence departures from it would be processed as perturbations, just as when the novel environment was first introduced. We based this hypothesis on previous work reporting that the removal of the novel condition is in-and-of itself a perturbation (Herzfeld et al., 2014b), and that feedback responses are adapted through experience to an altered environment even if there is no feedback-specific learning opportunities (Wagner and Smith, 2008; Yousif and Diedrichsen, 2012). During the split-to-tied transition in our experiment the leg that was 'fast' moves down in speed, while the one that was 'slow' moves up. This is the mirrored version of the initial tied-to-split transition where the 'fast' leg went up and the 'slow' leg went down. Accordingly, we expect the feedback-generated muscle activity to reflect this mirror-symmetry between belt-speed transitions when the split condition is introduced or removed. Formally expressed:

$$\Delta EMG_{split-to-tied} = eP_B - lA_B = eA_B^* \;\; \Rightarrow \;\; eP_B = lA_B + eA_B^*$$

Where eA_B^* is eA_B mirrored (i.e., transposing the values for the left and right legs).

2.5 *A general regression model to characterize the contribution of feedback-generated activity to aftereffects in muscle activity*

In summary, we tested our three hypotheses on the structure of feedback-generated components of aftereffects with one single regression model:

$$\Delta EMG_{split-to-tied} = eP_B - lA_B = eP_{lA} = -\beta_S eA_B + \beta_M eA_B^* \tag{2}$$

Where the parameters β_S and β_M are respectively interpreted as the extent of feedback-generated activity that is solely environment-dependent (β_S) or adaptive (β_M). Thus, the three hypotheses presented in the previous section can be summarized through the β coefficients as: (H1) negligible feedback contributions to aftereffects in muscle space ($\beta_S = \beta_M = 0$), (H2) solely environment-dependent (non-adaptive) feedback responses ($\beta_M = 0$ and $\beta_S = 1$), and (H3) adaptive feedback responses ($\beta_M = 1$ and $\beta_S = 0$).

It is worth pointing out that our regression model is considering the general case in which the initial muscle responses during early adaptation eA_B are not anti-symmetric (i.e., same group of muscles increasing in a leg, decrease in the other one). As a result the mirrored eA_B^* and opposite $-eA_B$ muscle vectors are hypothesized to be different from each other in muscle space (i.e., not colinear).

2.6 *Statistical analysis*

Differences in muscle activity across epochs. Significant changes in EMG variables between any pair of epochs were determined by using the Wilcoxon signed-rank test (i.e., non-parametric analogue of a paired t-test) on each of the 360 EMG parameters. Effect sizes were computed using median values across subjects since this is a measure less susceptible to outliers. All tests were two-tailed and the null hypothesis was that the (normalized) absolute effect size was smaller than 0.1 (i.e., 10% of the maximum baseline activity for that muscle). This was done to avoid finding significant but small, and presumably meaningless, differences. We corrected for multiple comparisons using the Benjamini-Hochberg procedure to control for False Discovery Rate (FDR) (Benjamini and Hochberg, 1995; Kass et al., 2016) for which we set an acceptable FDR to be 10%. We reasoned that this FDR value was adequate given our conservative approach of only considering significant effect sizes > 0.1. Threshold p-values from the FDR procedure are reported for each application.

Contribution of feedback activity to aftereffects. The linear regressions for testing our hypotheses on the structure of feedback-generated activity were performed using Matlab's *fitlm()* function and computing (Pearson's) R^2 values that were uncentered, given that our regression model did not include intercept terms.

Analysis of inter-subject variability. Lastly, we used Spearman's correlations to determine if either age or walking speed could explain the inter-subject variability in our model's coefficients. We focused on these subject-specific features because they exhibited large ranges in our cohort that could have impacted our results. Spearman's correlations were also used to study the association between aftereffects sizes in muscle activity and step-length asymmetry. For all these analyses we applied Spearman's correlations as a non-parametric alternative to Pearson's correlation because it is more robust to outliers. The correlation value (lowercase r, to distinguish from previous use of R) and the corresponding p-value were presented. To parse the relation between age and kinematic aftereffects onto muscle activity aftereffects, we used a two-factor linear regression. In this case there was no non-parametric method available, so we used a robust linear fit (bisquare weighting) to protect against potential outliers, and we report Pearson's R^2.

2.7 State-space modeling

We implemented a linear time-invariant state space model as an analytical tool, rather than a mechanistic model (e.g Thoroughman and Shadmehr, 1999), to characterize the evolution of EMG activity during the Adaptation and Post-Adaptation conditions. In particular, we used this framework to determine the extent to which the state of the system underlying the muscle activity during Adaptation predicted that of Post-Adaptation. To this end, we fitted a discrete linear time-invariant space-state model (Smith et al., 2006; Zarahn et al., 2008; Cheng and Sabes, 2006; Cheng and Sabes, 2007) using belt-speed state as input ($v_k = 0$ for tied-belts, 1 for split) and the averaged EMG activity across subjects (360 x 1 vector) at each stride k as output (y_k). Strides were considered to be the time indexes in the model. Equations for the model are:

$$x_{k+1} = Ax_k + Bv_k, \tag{3}$$

$$\hat{y}_k = Cx_k + Dv_k \tag{4}$$

Where $\hat{y}_k$ is the estimated EMG data and x_k is the state vector of the system at stride k. C and D are matrices

that weighted the effect of the states and input on the estimated data, respectively. The state vector x evolved according to the dynamics equation 3, where A is the matrix characterizing the temporal dynamics of the states and B weighed the effect of the input on the state evolution. We selected 2 states (size of x) in our model to directly compare our results to prior work using these number of states to characterize the adaptation dynamics of kinematic features in walking (Smith et al., 2006; Mawase et al., 2014). The initial states' value assumed to be $x = 0$. We assumed that the response of the system to a constant input (set by v_k) was described through the linear combination of decaying exponentials given the dynamics reported in prior models describing sensorimotor adaptation (Cheng and Sabes, 2006; Smith et al., 2006; Zarahn et al., 2008; Mawase et al., 2014). Thus, without loss of generality, we constrained the matrix A to be diagonal and the time-constants of the model were computed as $\tau_i = -1/log(a_{ii})$, where a_{ii} were the diagonal elements of the matrix A. The parameters A,B,C and D were fitted to the data using an E-M algorithm similar to that described in (Cheng and Sabes, 2006), custom written in Matlab R2017b (The Mathworks, Natick, MA, USA). The algorithm alternated between estimating of the matrices C and D, which were used to compute an estimation of the sequence x_k through least-squares. We then used the obtained sequence to estimate the matrices A and B that best fitted it in the least-squares sense. This in turn gives us a new estimate of x_k which can be used to re-estimate C and D. We initialized the matrix D by projecting the output data onto the (known) input vector v_k, and the columns of C as the first two (uncentered) principal components of the remainder $y_k - Dv_k$. We only used Adaptation data to fit the model parameters and assessed its prediction power over Postadaptation.

Previous uses of LTI-SSM models for description of motor adaptation have formulated the dynamical equation of the model in terms of an error signal (Smith et al., 2006; Zarahn et al., 2008; Mawase et al., 2014), rather than an input signal v_k as we do here. As long as the error signal is presumed to be a linear combination of motor output (y_k) and some external input (v_k), as is the case in all those examples, the models can be expressed in the form given by the equations above. However, a transformation of the parameters needs to be considered when comparing parameters across both formulations (Cheng and Sabes, 2007).

We also used the state-space approach described above to fit the EMG difference between legs at each stride k during Adaptation (i.e., $y_k = EMG_{sym} = EMG_{dominant} - EMG_{nondominant}$ = 180 x 1 vector) and predict EMG_{sym}

during Post-Adaptation. This was done given that EMG_sym would yield the same Post-Adaptation values, regardless of whether the structure of the feedback-generated activity was solely environment-dependent (H2) or adaptive (H3).

The goodness of fits and prediction were quantified through the RMS residuals: $e = k\frac{\sum_{i=1}^{N}\sqrt{\sum_j (y_{ij}-\hat{y}_{ij})^2}}{N}$, where y_{ij} represents the data for the j-th EMG variable (e.g., $j \in [1,360]$ for bilateral EMG fitting) during the i-th stride, $\hat{y}_{ij}$ is the corresponding model output, and N is the number of strides considered. k is a model-specific constant that normalizes residuals such that $e = 1$ for the Baseline epoch, to facilitate comparisons across models. Residuals are presented for both models in each of the main analysis epochs (eA, lA, eP). For comparison, residuals are also presented for each epoch with respect to that epoch's mean behavior (i.e. residuals due to trial-to-trial variability). For display purposes of each model's goodness of fit, we projected the empirical data to the subspace spanned by the columns of the matrix C and the matrix D. This allows us to visualize differences between the expected evolution of each state (given by Equation 3), and the data constrained to the relevant subspaces given by the model. Finally, we tested if the same dynamics (i.e. time-constants) describing EMG data were able to describe step-length asymmetry (SLA) data. We did this by taking the states fitted to EMG data (i.e. preserving the fitted parameters of Equation 3) but we used SLA data as the output for Equation 4. This required us to fit a new 2×1 matrix C_{SLA} and a scalar D_{SLA}.

3 Results

3.1 Muscle activity during early Adaptation reflects feedback control

Muscle activity when the split condition was introduced primarily reflected feedback-generated motor commands. This is shown by the changes in body position Figure2A and muscle activity Figure2C during Early Adaptation compared to Baseline walking (i.e., eA_B). We observed that the body position (i.e., hip position) with respect to the standing foot during stance phase (e.g., fast leg's foot for the fast stance) was either farther in front (positive values) during fast stance or farther back (negative values) during slow stance in Early Adaptation compared to Baseline walking (Figure2A). In other words, subjects were either "falling" forward or backward during Early Adaptation immediately after hitting the ground (heel-strike) with the fast or slow legs, respectively. This was likely due to the discrepancy between subjects' expected and actual walking speed (Figure2B). Consistently, the corresponding muscle activity for both legs during 100-400ms (colored rectangles) after heel-strike (onset of movement perturbation at each stride) resembled those of feedback-mediated postural responses over comparable time windows (Torres-Oviedo and Ting, 2007; Chvatal and Ting, 2012; Safavynia and Ting, 2012; De Kam et al., 2017). Namely, the standing fast leg following fast heel-strike (blue rectangle) initially increased the activity of posterior muscles in the ankle (MG, LG, SOL) and knee (SMB, SMT, BF) followed by increased activity in quads (VM,VL,RF) and hip flexors (ADM,HIP, TFL) (Figure 2C), which has been previously observed when the body falls forward (Chvatal et al., 2011; Chvatal and Ting, 2012). Concurrently, the slow leg exhibited greater activity of hamstrings (SMB, SMT, BF), ankle (TA) and hip flexors (RF, ADM, HIP) to presumably swing the leg more vigorously as a possible feedback-mediated stepping balance response (Horak and Macpherson, 1996). Similarly, the standing slow leg following the slow heel-strike (orange rectangle) initially increased the activity of anterior muscles (TA, quads -RF, VM, and VL-, and hip flexors -HIP,ADM,TFL-) and reduced the activity of calf muscles (MG, LG, SOL) (Figure 2C) as has been observed in balance responses when the body falls backward (Tang and Woollacott, 1999; Chvatal et al., 2011; Chvatal and Ting, 2012). On the other hand, some activity during Early Adaptation might have been modulated according to motor demands that persist throughout the split-belt condition. Particularly, the fast leg's swing activity (increased TA, hamstrings -SMB, SMT, and BF-, and hip flexors -ADM and HIP-) and the activity of ankle muscles (MG, LG, SOL, PER) during later phase of stance for both legs were also observed during

Late Adaptation (Figure3B). In sum, most of the changes observed during Early Adaptation matched those of feedback-mediated responses to counteract balance perturbations induced by the novel environment.

3.2 Anti-symmetric modulation of muscle activity during Late Adaptation is not a direct reflection of each leg's walking speed

Muscle activity during Late Adaptation was not simply regulated as a function of the speed at which each leg moved in the split condition. Overall we observed an anti-symmetric modulation of muscle activity of the two legs during Late Adaptation with respect to Baseline (lA_B displayed in Figure 3B). Namely, we found that if a group of muscles increased activity on one side, the same group decreased activity on the other one. Interestingly, this opposing modulation across legs was not merely determined by ipsilateral walking speed. Should this have been the case, one would expect reduced muscle activity of the slow leg with respect to mid-speed Baseline as seen in slow walking (Figure 3A) and vice versa for the fast leg (Den Otter et al., 2004; Dietz et al., 1994). However, this type of modulation was only observed in distal muscles (MG, LG, SOL, PER, TA) and in the fast leg's hip flexors (HIP, TFL, ADM), but not in most of the proximal muscles (SMB, SMT, BF, RF, VM, VL, ADM, TFL). In fact, proximal muscles crossing the knee (SMB, SMT, BF, RF, VM, VL) and the slow leg's hip (ADM, TFL) increased in the leg walking slow and decreased in the leg walking fast. This suggested that their activity was predominantly regulated by the speed of the contralateral side. This was least anticipated in quads (RF, VM, VL) and hip flexors (ADM, TFL) of the standing leg since their activation was opposite to that of other muscles within the same leg. In contrast, the modulation of hamstrings (SMB, SMT, BF) during swing was less unexpected since it matched concurrent modulations of the contralateral standing leg. In sum, muscle activity for split-belt walking highlights the interlimb nature of locomotion because it exhibited an interaction between muscle group (i.e., distal vs. proximal) and walking speed, rather than individual leg modulation based on speed.

3.3 Feedback responses are transposed across legs when returning to tied-belts after prolonged exposure to split-belts

We proposed that aftereffects (eP_B) would be generated by a combination of feedforward and feedback processes. In particular, one of our hypotheses (H1) predicted that feedforward-generated motor commands would dominate EMG aftereffects, such that muscle activity during Early Post-Adaptation would be similar to that of Late Adaptation (lA_B). Qualitatively, we found little resemblance in the muscle activity during these two epochs, as shown by the few significant changes that were similar between them (black outlines in Figure 3C). This indicated that some form of feedback-generated motor commands also contributed to muscle patterns during Early Post-Adaptation. To quantitatively rule out H1 and distinguish between the two remaining hypotheses (H2 and H3), we used a regression model ($eP_{lA} = -\beta_S eA_B + \beta_M eA_B^*$) to contrast feedback-generated activity upon introduction (eA_B) and removal (eP_{lA}) of the split perturbation (Figure4). Recall that H2 predicted these muscle patterns would be numerically opposite (i.e., $eP_{lA} = -\beta_S eA_B$, with $\beta_S = 1$), whereas H3 predicted they would mirror each other (i.e., $eP_{lA} = \beta_M eA_B*$, with $\beta_M = 1$ and eA_B* a mirrored version of eA_B, such that the slow leg's activity in one transition would exactly be the fast leg's activity in the other transition and vice versa). On the other hand, H1 predicted feedback-mediated activity would be negligible compared to feedforward one (i.e., $\beta_S = 0$ and $\beta_M = 0$).

Our regression analyses supported the mirroring hypothesis (H3) when using group averaged activity (Figure 4E, large magenta dot: CI for $\beta_M = [.693, .782]$, $\beta_S = [.156, .244]$, $R^2 = 0.753$) or muscle activity for each individual (Figure 4E, small magenta dots; median $\pm$ inter-quartile range for $\beta_M = 0.442 \pm 0.177$, $\beta_S = 0.193 \pm 0.396$, and $R^2 = 0.321 \pm 0.258$). Qualitatively, one can observe that the pattern of the slow leg at tied-to-split transition (Fig. 4C, top half) was remarkably similar to the one of the fast leg at split-to-tied transition (Fig. 4D, bottom half). This mirroring was also observed when comparing the contralateral sides (Fig. 4C bottom half to 4D top half) at these transitions, except for the reduced activity in some muscles during early stance (RF, VL, VM, GLU, TFL). We also found that the body motion exhibited mirroring between the legs' stance phases. For example, the change in body position with respect to the fast stance leg in the tied-to-split transition (Fig. 4F, green trace) closely resembled the one during slow stance at the split-to-tied transition (Fig. 4F, magenta

trace). Thus, both EMG and kinematics exhibited mirroring when we consider changes with respect to the previous steady-state condition. This is consistent with the nervous system (specifically, the feedback response mechanisms) adopting the steady-state behavior as a new reference gait pattern. Perturbation of kinematics away from this new reference, and not perturbations away from the subjects' baseline behavior, may drive feedback-generated muscle activity.

To confirm that the mirroring between legs is a learning-dependent process, and not simply due to removal of the split perturbation, we ran the same regressions on the split-to-tied transition following the Short Exposure condition. In this condition subjects did not have time to adapt, so we expected to observe solely environment-dependent changes in EMG activity (H2), rather than mirroring (H3). Indeed, that is what we found with our regression model applied the transition after the Short Exposure. This was true for both the group-averaged activity (Figure 4E, large gray dot; CI for $\beta_M = [-.0671, .0262]$, $\beta_S = [1.16, 1.25]$, $R^2 = 0.881$) and the activity of individual subjects (Figure 4E, small gray dots; median $\pm$ inter-quartile range for $\beta_M = 0.0624 \pm 0.355$, for $\beta_S = 0.911 \pm 0.471$, and for $R^2 = 0.612 \pm 0.102$). These results show a strong dissociation of the structure of change in feedback responses and exposure duration: following the Short Exposure condition, feedback muscle activity can be modeled as environment-dependent, whereas following Adaptation (long exposure) the feedback is well explained by mirroring. Taken together, we found that feedback-generated activity was adapted and strongly contributed to the structure of aftereffects in muscle space.

3.4 Healthy aging reduces the adaptation of feedback-generated muscle activity contributing to aftereffects

Analysis of intersubject variability revealed that older adults exhibited less adaptation of feedback-generated activity. Namely, β_M and β_S were associated to subjects' age ($r = -0.64$, $p = 0.0098$ and $r = 0.59$, $p = 0.017$ respectively, Figure 5A) with older subjects showing smaller β_M and larger β_S. This indicates that feedback activity in older adults was less adapted (and more environment-driven) compared to younger adults. We also noticed that the magnitude of feedback activity were possibly smaller in older adults ($\|\Delta EMG_{split-to-tied}\| = \|eA_B\|$: $r = -0.54$, $p = 0.032$; $\|\Delta EMG_{tied-to-split}\| = \|eP_B - lA_B\|$: $r = -0.47$, $p = 0.07$; Figure 5B). These smaller feedback responses in older individuals could make it more difficult to identify the structure of feedback

activity because of the reduced signal to noise ratio, leading to biased or noisy estimates of β_M and β_S. To discard this possibility, we correlated the R^2 of the fitted models with age. We found no effects of age ($r = -0.36$, $p = 0.17$, not shown), meaning that the regression model applied to individual data captured comparable levels of variance regardless of subject age. Taken together, we observed an actual age-mediated difference in the extent of mirroring of feedback muscle activity, suggesting an age-mediated decline of learning processes updating feedback responses. This limited learning capacity in older individuals was further supported by the negative association between age and EMG aftereffects ($\|eP_B\|$, $r = -0.67$, $p = 0.0054$ Figure 5C): the older the subjects, the smaller the aftereffects. This happened despite the similar magnitude of muscle activity during late Adaptation across individuals ($\|lA_B\|$ vs. age: $r = 0.25$, $p = 0.35$, Figure 5C). Importantly, walking speed, which naturally alters muscle activity, was not associated to either the magnitude of EMG aftereffects ($p = 0.36$) nor to the adaptation (speed vs. betaM $p = 0.64$) or magnitude of split-to-tied feedback responses ($p = 0.59$). Interestingly, no age mediation was observed in the magnitude of step-length asymmetry aftereffects ($r = -0.16$, $p = 0.55$, Figure 5D), which are conventionally used to characterize locomotor learning. This suggests that the adaptation of muscle activity may be more sensitive to individual differences in learning than kinematics. Alternatively, there might be partially different mechanisms involved in the adaptation of muscle activity and step length asymmetry. In agreement, muscle activity aftereffects and kinematic aftereffects were not correlated ($r = 0.38$, $p = 0.15$, not shown) unless the effect of age on EMG aftereffects is accounted for. Specifically, the robust linear regression (see Methods) of muscle activity aftereffects onto both kinematic aftereffects and age indicated that both factors were significant predictors (Pearson's $R^2 = 0.741$, $p_{age} = 0.018$ and $p_{SLA} = 4.2 \times 10^{-4}$, not shown). We interpret these results as supportive of two different learning mechanisms: one that underlies the association between age and EMG aftereffects, and a separate one that is captured by the association between EMG and kinematics aftereffects. In sum, we observed diminished mirroring of feedback activity post-adaptation and smaller EMG aftereffects in older adults, suggesting that healthy aging has a negative impact on learning process updating feedback-generated muscle activity.

3.5 *The dynamics of Adaptation predicts Post-Adaptation of EMG symmetry, but not of individual legs*

The adaptive nature of feedback-generated activity (Hypothesis H3 in Figure 1C) was further supported by the poor prediction of EMG aftereffects of individual legs compared to symmetry measures (Figure 6). Recall that only purely environment-driven feedback responses (Hypothesis H2 in Figure 1C) can be reproduced by a linear model framework. In other words, a necessary condition for a system to be linear time-invariant (LTI) is that its response (output) upon introduction of a given perturbation (input) would be numerically opposite to the one observed upon removal of said perturbation after reaching steady state. Consistently, our state-space model predicted that any muscle whose activity increased when the split perturbation was introduced during Adaptation, should decrease by the same amount when the perturbation is removed during Post-Adaptation (and vice versa) (Simulation on Figure 6E). This was not what we observed (see sections 3.3 and 3.4). Thus, the state-space framework poorly reproduced the EMG activity during Post-Adaptation (normalized mean residual $e = 4.43$ for eP_B) despite the relatively good fit of EMG variables during Adaptation ($e = 1.58$ for eA_B and $e = 1.11$ for lA_B) relative to the residuals around the mean of the actual data ($e = 1.55$ for eP_B, $e = 1.53$ for eA_B, and $e = 1.05$ for lA_B) (Figure 6E).

In contrast, an LTI state-space model was sufficient to fit the Adaptation (data $e = 1.57$ vs. simulated $e = 1.64$ for eA_B; data $e = 1.13$ vs. simulated $e = 1.16$ for lA_B) and predict the aftereffects of EMG asymmetry (data $e = 1.48$ vs. simulated $e = 2.31$ for eP_B). This was because EMG asymmetry (difference between legs) collapsed each legs' muscle activity to a subspace for which mirrored (H3) and numerically opposite (H2) responses were equivalent. From a practical perspective, the estimated time constants for the adaptation of EMG asymmetry were $\tau_1 = 39.5$ and $\tau_2 = 632$ strides. Remarkably, the time constants extracted from EMG data during Adaptation reproduced well the evolution of step length asymmetry (Figure 6 bottom row), but to a lesser extent the evolution of EMG asymmetry during Post-Adaptation. Namely, only the EMG data decaying faster was well predicted. This is shown by the overlap between simulated (solid line) and Post-Adaptation data (blue dots) decaying faster, but the mismatch for the data de-adapting slower (orange dots). Further studies are needed to investigate this discrepancy. In summary, LTI-state space models solely based on Adaptation data can predict some of the evolution of asymmetry measures during Post-Adaptation, but not of individual legs.

4 Discussion

We have shown that aftereffects following split-belt walking were not merely a continuation of adapted muscle activity patterns, but they exhibited a large contribution of feedback-generated motor commands. Further, we showed that the elicited feedback responses were adapted, such that removing the split condition induced muscle activity similar to the one observed when unexpectedly increasing or decreasing walking speed upon introduction of the perturbed environment. This indicates that subjects adopted the split pattern as their reference gait, which cannot be modeled with a linear system predicting that feedback motor commands would be reversed when the split environment was removed. Interestingly, this phenomenon was age-dependent, with older subjects showing less adapted feedback commands leading to smaller aftereffects in muscle space. Our results have implications for the design of rehabilitation treatments using split-belt treadmills because we provide a full characterization of expected changes in muscle activity during and after split-belt walking for age ranges similar to those of post-stroke individuals.

Aftereffects in muscle activity are not a continuation of late adaptation activity.

We have shown that aftereffects following split-belt walking were not merely a continuation of adapted muscle activity patterns. Notably, only a few muscles exhibited similar changes during late Adaptation and early Post-Adaptation. We expected to find more similarities between these two epochs given that kinematic aftereffects are considered to mostly reflect a continuation of motor commands adapted in the altered environment (Malone et al., 2012; Morton and Bastian, 2006; Ogawa et al., 2014). Plantarflexors during terminal stance (i.e., during the second double support phase) were some of the few muscles that showed continuity when the split perturbation was removed. They specifically increased activity in the leg walking fast and decreased activity in the leg walking slow. These muscles generate the propulsion force to displace the trunk and swing the leg (Neptune et al., 2001; Neptune et al., 2008) and hence modulate step length (Neptune et al., 2008). These muscles increased activity in one leg and decreased activity in the other. Thus, the plantarflexor activity during Adaptation might be regulated to equalize step lengths in the split environment and therefore its continuation upon returning to tied walking may drive step-length asymmetry aftereffects. To test this hypothesis and fully characterize the dependency of kinematic aftereffects on muscle activity Post-Adaptation requires future work

with a distinct approach, such as simulations with models of the musculo-skeletal system (Song and Geyer, 2015; Steele et al., 2010)

The structure of feedback activity contributing to aftereffects reflects the adaptation of internal model for gait.

While feedback activity is expected upon a sudden change in the environment, we observed that feedback responses were not simply reflecting the environment-dependent change. This was indicated by the difference in feedback activity after identical transitions in the environment following distinct exposures of the split condition. Notably, we observed mirroring between the legs only after a long-exposure of split-belt walking, suggesting that this was a learning-dependent effect. In other words, each leg exhibited feedback activity upon removal of the split condition that was similar to the one experienced by the other leg upon introduction of the novel situation. This mirroring in feedback motor commands is consistent with the observation that removal of a sustained perturbation facilitates learning of the opposite perturbation because of the similarity of errors elicited by both situations (Herzfeld et al., 2014b; Roemmich and Bastian, 2015). Adaptation of feedback responses has been observed in reaching too (Wagner and Smith, 2008; Yousif and Diedrichsen, 2012; Crevecoeur and Scott, 2013; Cluff and Scott, 2013), but this has not been systematically tested in walking. In sum, feedback-generated motor commands contribute to aftereffects and their structure indicate that they are adapted.

We consider three possibilities on how the the feedback loop may be adapted. First, the feedback policy transforming the sensed state into corrective actions might rely on on internal models that are adapted through exposure to an alternative environment (Bhushan and Shadmehr, 1999). Second, the state estimation may be inaccurate due to sensory shifts following split-belt walking (Vazquez et al., 2015). As a result, feedback commands might be different post-adaptation even if the feedback policy remains unchanged. Third, feedback adaptation might be merely a byproduct of feedforward adaptation. That is, feedback activity post-adaptation might be generated because of a misapplied motor pattern, arguably generated through a feedforward mechanism. While possible, we believe this last option cannot fully explain our observations given that age-related decay is

observed in the adaptation (i.e., mirroring) of feedback activity but not in kinematic aftereffects.

Neural substrates underlying the adaptation of feedback activity.

Feedforward adaptation and learning of internal models is known to rely on cerebellar function (Wolpert et al., 1998; Morton and Bastian, 2006; Galea et al., 2011; Therrien and Bastian, 2015). Thus, the cerebellum may also be required for adaptation of feedback responses. Indeed, it has been observed that the cerebellum is required for timely recruitment of feedback responses (Herzfeld et al., 2014a). Primary motor cortex (M1) has been linked to retention of adapted states (Galea et al., 2011). Feedback responses, even non-voluntary ones, have also been shown be causally mediated by M1 (Pruszynski et al., 2016) during reaching. More specifically to our case, postural responses are thought to be recruited by basal ganglia circuitry, and are modulated by prior experience through cortical input (Jacobs and Horak, 2007; Bolton, 2015). Further, patients with cerebellar lesions cannot modulate the strength of their responses to predictable perturbations (Jacobs and Horak, 2007) It is plausible then that feedback and feedforward responses may share an adaptive internal model (Wagner and Smith, 2008; **?**), which relies on the cerebellum for its adaptation, and on M1 for its later influence on movement.

Age modulates feedback response adaptation

Aging had a distinct effect on the adaptation of muscle activity during and after split-belt walking, suggesting a possible dissociation on the execution or update of feedback and feedforward activity. Motor learning affected by age as indicated by the reduced magnitude of aftereffects and less mirroring in feedback activity post-adaptation. It has been suggested that older subjects have less reliable sensory information (Konczak et al., 2012), and thus reduce their sensory information weighting and rely more on predictive mechanisms (Wolpe et al., 2016). This would explain both the weaker feedback responses upon environmental transitions ($\|eA_B\|$ and $\|eP_B - lA_B\|$) and reduced recalibration of internal models (β_M) in older subjects, consistent with our observations. Despite the age modulation of muscle activity aftereffects, no age dependence in kinematic aftereffects (SLA) was observed. These kinematic observations are consistent with previous studies on aging in split-belt walking (Sombric et al., 2017; Bruijn et al., 2012). It may just be that EMG signals, being a closer

correlate of neural activity than kinematics, are more sensitive to differences in internal model recalibration (Thoroughman and Shadmehr, 1999). Alternatively, this EMG/kinematic discrepancy on age-related modulation of aftereffects may imply a partial dissociation of the underlying neural circuitry of each type of aftereffect. Further, the magnitude of muscle patterns during Late Adaptation, which is mostly feedforward-generated activity, was not affected with age. The dichotomy in age effects on feedback vs. feedforward activity suggests a partial distinction in the neural mechanism underlying the adaptation or execution of feedback and feedforward motor commands. Dissociation of feedback and feedforward adaptation has been observed through training on inconsistent environments (Yousif and Diedrichsen, 2012). Thus, it is possible that both types of adaptation are only partially overlapping in their substrates. Further work should try to dissociate these processes for split-belt walking. We conclude that older subjects rely less on sensory information, but this becomes evident only by looking beyond kinematic measures of task performance.

Limitation of LTI state space frameworks when considering the evolution of individual legs. Linear time-invariant models have been used profusely to characterize the temporal dynamics of motor adaptation (Smith et al., 2006; Cheng and Sabes, 2007; Zarahn et al., 2008). This is particularly important when used as a prediction tool. For example, state-space model can predict the impact of different adaptation protocols on the post-adaptation dynamics (Joiner and Smith, 2008). We found remarkable that muscle activity during adaptation could partially predict the activation patterns of de-adaptation without condition-dependent parameter fitting (Mawase et al., 2014) or data scaling (Roemmich et al., 2016). However, we could only predict Post-Adaptation muscle patterns from Adaptation dynamics of asymmetry measures and not of individual legs. Notice that this issue is independent of other limitations that LTI models may have, such as their inability to describe savings (Zarahn et al., 2008). This limitation is worth noting given the clinical interest to predict the effect of distinct adaptation protocols on the paretic side, rather than on the relative differences between the legs. In sum, the recalibration of the internal model underlying the adaptation of gait can only be represented by an LTI state-space model when considering the evolution of asymmetry measures, but not of individual legs.

Changes in muscle activity through Adaptation The steady-state pattern of muscle activity in the split-belts

environment (late Adaptation) is not consistent with simple ipsilateral speed-based modulation of activity. Our observation is that distinct muscle groups up- and down-regulate their activity on both legs, whereas ipsilateral speed-based modulation would predict down-regulation only in the slow leg, and up-regulation on the fast one. This highlights the bilateral coordination requirements of walking, and suggests the adapted state of the two legs are not decoupled from one another. In contradiction to our finding, it has been suggested that adaptation to split-belt walking may happen through leg-specific neural substrates that can be adapted independently (Choi and Bastian, 2007). We hypothesize that coupling is likely dependent on the constraints between the legs imposed by the environment, which would explain the difference in results: (Choi and Bastian, 2007) used hybrid walking, with one leg walking forward and the other backward, a situation that may be more easily decoupled into independent patterns.

How does the nervous system converge to the new steady-state (late Adaptation) pattern of activity? One possibility is that early feedback activity may act as a tutor for the neural circuits responsible for learning a new feedforward pattern (Thoroughman and Shadmehr, 1999; Albert and Shadmehr, 2016). Consistent with this idea, early and late Adaptation activity are qualitatively similar on most muscle groups. However, many muscles increase their activity during early Adaptation but end up showing no modulation during late Adaptation, and even those that do end up having smaller levels of activity. This is, activity in most muscles and phases of the gait cycle is reduced from early to late Adaptation. This coincides with observations showing muscle activity is strictly decreasing during adaptation to some perturbed environment and a corresponding metabolic cost reduction (Huang and Ahmed, 2014; Finley et al., 2013), and is consistent with a model where initial muscle activity modulation is aimed at improving performance while simultaneously increasing stability (e.g. through co-contraction) in the presence of new dynamics, and is gradually reduced until settling on a more efficient but equally useful pattern of activity (Franklin et al., 2008).

Clinical implications Split-belt treadmills have been suggested as a potentially effective treatment in the recovery of gait symmetry in the chronic phase (Reisman et al., 2007; Reisman et al., 2013). The systematic description of healthy subjects' muscle activity (normative data) presented here will allow for a better characterization of stroke subjects' departure from this behavior and paves the way to performing early diagnosis of the therapeutic

potential of the split-belt treadmill. Previous reports on muscle activity during locomotor adaptation have looked at reduced muscle sets (Ogawa et al., 2014; Dietz et al., 1994), focused on coordination across muscles (Maclellan et al., 2014), or reported results for only portions of the gait cycle or activity averaged throughout the full cycle (Raja et al., 2013; Dietz et al., 1994). Importantly, we present amplitude of muscle activity during distinct phases of the gait cycle, as opposed to integrated muscle activity during such phases. Timing of the gait cycle is altered during Adaptation and presents some aftereffects (Reisman et al., 2005), which confounds the origin of observed changes in time-integrated EMG.

Within our findings, three elements appear of importance towards the development of personalized rehabilitation treatments. First, the age dependence of some responses suggests that patient age needs to be considered a potential factor when assessing the effectiveness of treatment. Second, assessment of learning through kinematic-only measurements may be missing interesting insights into neural activity changes which may be better captured through EMG-based measurements. Third, aftereffects following a sudden change in walking dynamics may be more strongly influenced by the feedback reaction to that change than by changes in feedforward circuits, which are arguably the true target of rehabilitation therapies. Thus, long-term rehabilitative potential may not be well captured by measuring behavior immediately after split-belts treadmill walking.

Albert ST, Shadmehr R (2016) The Neural Feedback Response to Error As a Teaching Signal for the Motor Learning System. *Journal of Neuroscience* 36:4832–4845.

Benjamini Y, Hochberg Y (1995) Controlling the false discovery rate: a practical and powerful approach to multiple testing.

Bernstein N (1967) *The co-ordination and regulation of movements* Pergamon Press.

Bhushan N, Shadmehr R (1999) Computational nature of human adaptive control during learning of reaching movements in force fields. *Biological cybernetics* 81:39–60.

Bolton DAE (2015) The role of the cerebral cortex in postural responses to externally induced perturbations. *Neuroscience and Biobehavioral Reviews* 57:142–155.

Bruijn SM, Van Impe A, Duysens J, Swinnen SP (2012) Split-belt walking: adaptation differences between young and older adults. *Journal of Neurophysiology* 108:1149–1157.

Cheng S, Sabes PN (2006) Modeling Sensorimotor Learning with Linear Dynamical Systems. *Neural Computation* 18:760–793.

Cheng S, Sabes PN (2007) Calibration of visually-guided reaching is driven by error corrective learning and internal dynamics. *Journal of neurophysiology* 97:3057–3069.

Choi JT, Bastian AJ (2007) Adaptation reveals independent control networks for human walking. *Nature Neuroscience* 10:1055–1062.

Chvatal SA, Ting LH (2012) Voluntary and Reactive Recruitment of Locomotor Muscle Synergies during Perturbed Walking. *The Journal of Neuroscience* 32:12237–12250.

Chvatal SA, Torres-Oviedo G, Safavynia SA, Ting LH (2011) Common muscle synergies for control of center of mass and force in nonstepping and stepping postural behaviors. *Journal of neurophysiology* 106:999–1015.

Cluff T, Scott SH (2013) Rapid Feedback Responses Correlate with Reach Adaptation and Properties of Novel Upper Limb Loads. *Journal of Neuroscience* 33:15903–15914.

Crevecoeur F, Scott SH (2013) Priors Engaged in Long-Latency Responses to Mechanical Perturbations Suggest a Rapid Update in State Estimation. *PLoS Computational Biology* 9.

De Kam D, Roelofs JM, Bruijnes AK, Geurts AC, Weerdesteyn V (2017) The Next Step in Understanding Impaired Reactive Balance Control in People with Stroke: The Role of Defective Early Automatic Postural Responses. *Neurorehabilitation and Neural Repair* 31:708–716.

Den Otter AR, Geurts ACH, Mulder T, Duysens J (2004) Speed related changes in muscle activity from normal to very slow walking speeds. *Gait and Posture* 19:270–278.

Dietz V, Zijlstra W, Duysens J (1994) Human neuronal interlimb coordination during split-belt locomotion .

Finley JM, Bastian AJ, Gottschall J (2013) Learning to be economical: the energy cost of walking tracks motor adaptation. *The Journal of Physiology* 591:1081–1095.

Franklin DW, Burdet E, Peng Tee K, Osu R, Chew CM, Milner TE, Kawato M (2008) CNS Learns Stable, Accurate, and Efficient Movements Using a Simple Algorithm. *Journal of Neuroscience* 28:11165–11173.

Galea JM, Vazquez A, Pasricha N, Orban de Xivry JJ, Celnik P (2011) Dissociating the Roles of the Cerebellum and Motor Cortex during Adaptive Learning: The Motor Cortex Retains What the Cerebellum Learns. *Cerebral Cortex* 21:1761–1770.

Herzfeld DJ, Pastor D, Haith AM, Rossetti Y, Shadmehr R, O'Shea J (2014a) Contributions of the cerebellum and the motor cortex to acquisition and retention of motor memories. *NeuroImage* 98:147–158.

Herzfeld DJ, Vaswani Pa, Marko M, Shadmehr R (2014b) A memory of errors in sensorimotor learning - Supplementary Materials. *Science (New York, N.Y.)* pp. 1–30.

Horak FB, Macpherson JM (1996) Postural orientation and equlibrium In *Handbook of Physiology. Excercise: Regulation and Integration of Multiple Systems.*, chapter 7, pp. 255–292. American Physiological Society, Bethesda, MD.

Horak FB, Nashner LM, Diener HC (1990) Postural strategies associated with somatosensory and vestibular loss. *Experimental Brain Research* 82:167–177.

Huang HJ, Ahmed AA (2014) Reductions in muscle coactivation and metabolic cost during visuomotor adaptation. *Journal of Neurophysiology* 112:2264–2274.

Jacobs JV, Horak FB (2007) Cortical control of postural responses. *Journal of Neural Transmission* 114:1339–1348.

Jordan MI, Rumelhart DE (1992) Forward models: supervised learning with a distal teacher. *Cognitive Science* 307354:16:307–354.

Kass RE, Caffo BS, Davidian M, Meng XL, Yu B, Reid N (2016) Ten Simple Rules for Effective Statistical Practice. *PLoS Computational Biology* 12.

Konczak J, Sciutti A, Avanzino L, Squeri V, Gori M, Masia L, Abbruzzese G, Sandini G (2012) Parkinson's disease accelerates age-related decline in haptic perception by altering somatosensory integration. *Brain* 135:3371–3379.

Lewek MD, Braun CH, Wutzke C, Giuliani C (2017) The role of movement errors in modifying spatiotemporal gait asymmetry post stroke: a randomized controlled trial. *Clinical Rehabilitation* p. 026921551772305.

Maclellan MJ, Ivanenko YP, Massaad F, Bruijn SM, Duysens J, Lacquaniti F (2014) Muscle activation patterns are bilaterally linked during split-belt treadmill walking in humans. *Journal of neurophysiology* 111:1541–52.

Malone LA, Bastian AJ, Torres-Oviedo G (2012) How does the motor system correct for errors in time and space during locomotor adaptation? *Journal of neurophysiology* 108:672–83.

Mawase F, Shmuelof L, Bar-Haim S, Karniel A (2014) Savings in locomotor adaptation explained by changes in learning parameters following initial adaptation. *Journal of neurophysiology* 111:1444–54.

Merletti R, Parker P (2005) *Electromyography*.

Morton SM, Bastian AJ (2006) Cerebellar contributions to locomotor adaptations during splitbelt treadmill walking. *The Journal of Neuroscience* 26:9107–16.

Mozaffarian D, Benjamin EJ, Go AS, Arnett DK, Blaha MJ, Cushman M, Das SR, Ferranti SD, Després JP, Fullerton HJ, Howard VJ, Huffman MD, Isasi CR, Jiménez MC, Judd SE, Kissela BM, Lichtman JH, Lisabeth LD, Liu S, MacKey RH, Magid DJ, McGuire DK, Mohler ER, Moy CS, Muntner P, Mussolino ME, Nasir K, Neumar RW, Nichol G, Palaniappan L, Pandey DK, Reeves MJ, Rodriguez CJ, Rosamond W, Sorlie PD,

Stein J, Towfighi A, Turan TN, Virani SS, Woo D, Yeh RW, Turner MB (2016) *Heart disease and stroke statistics-2016 update a report from the American Heart Association*, Vol. 133.

Neptune RR, Kautz SA, Zajac FE (2001) Contributions of the individual ankle plantar flexors to support, forward progression and swing initiation during walking. *Journal of Biomechanics* 34:1387–1398.

Neptune RR, Sasaki K, Kautz SA (2008) The effect of walking speed on muscle function and mechanical energetics. *Gait and Posture* 28:135–143.

Ogawa T, Kawashima N, Ogata T, Nakazawa K (2014) Predictive control of ankle stiffness at heel contact is a key element of locomotor adaptation during split-belt treadmill walking in humans. *Journal of neurophysiology* 111:722–32.

Perry J, Burnfield JM (2010) *Gait Analysis Normal and Pathological Function* Slack Inc., 2nd edition.

Pruszynski JA, Kurtzer IL, Nashed JY, Omrani M, Brouwer B, Scott SH (2016) Primary motor cortex underlies multi-joint integration for fast feedback control. *Nature* 478:387–390.

Raja B, Neptune RR, Kautz SA (2013) Coordination of the non-paretic leg during hemiparetic gait: expected and novel compensatory patterns. *Clinical biomechanics (Bristol, Avon)* 27:1023–1030.

Reisman DS, Block HJ, Bastian AJ (2005) Interlimb coordination during locomotion: what can be adapted and stored? *Journal of neurophysiology* 94:2403–15.

Reisman DS, McLean H, Keller J, Danks KA, Bastian AJ (2013) Repeated split-belt treadmill training improves poststroke step length asymmetry. *Neurorehabilitation and neural repair* 27:460–8.

Reisman DS, Wityk R, Silver K, Bastian AJ (2007) Locomotor adaptation on a split-belt treadmill can improve walking symmetry post-stroke. *Brain : a journal of neurology* 130:1861–72.

Rikli RE, Jones CJ (1998) The Reliability and Validity of a 6-Minute Walk Test as a Measure of Physical Endurance in Older Adults.

Roemmich RT, Bastian AJ (2015) Two ways to save a newly learned motor pattern. *Journal of Neurophysiology* 113:3519–3530.

Roemmich R, Long A, Bastian AJ (2016) Seeing the Errors You Feel Enhances Locomotor Performance but Not Learning. *Current Biology* 26:2707–2716.

Safavynia SA, Ting LH (2012) Task-level feedback can explain temporal recruitment of spatially fixed muscle synergies throughout postural perturbations. *Journal of neurophysiology* 107:159–77.

Sánchez N, Park S, Finley JM (2017) Evidence of Energetic Optimization during Adaptation Differs for Metabolic, Mechanical, and Perceptual Estimates of Energetic Cost. *Scientific Reports* 7:7682.

Scheidt RA, Reinkensmeyer DJ, Conditt MA, Rymer WZ, Mussa-Ivaldi FA (2000) Persistence of motor adaptation during constrained, multi-joint, arm movements. *Journal of neurophysiology* 84:853–862.

Smith MA, Ghazizadeh A, Shadmehr R (2006) Interacting adaptive processes with different timescales underlie short-term motor learning. *PLoS biology* 4:e179.

Sombric C, Calvert JS, Torres-Oviedo G (2018) Large propulsion demands increase locomotor learning at the expense of step length asymmetry. *bioRxiv* p. 372425.

Sombric CJ, Harker HM, Sparto PJ, Torres-Oviedo G (2017) Explicit Action Switching Interferes with the Context-Specificity of Motor Memories in Older Adults. *Frontiers in Aging Neuroscience* 9.

Song S, Geyer H (2015) A neural circuitry that emphasizes spinal feedback generates diverse behaviours of human locomotion. *Journal of Physiology* 593:3493–3511.

Steele KM, Seth A, Hicks JL, Schwartz MS, Delp SL (2010) Muscle contributions to support and progression during single-limb stance in crouch gait. *Journal of Biomechanics* 43:2099–2105.

Tang P, Woollacott MH (1999) Phase-depdendent modulation of proximal and distal postural responses to slips in young and older adults. *Journal of Gerontology: Medical Science* 54:M89–M102.

Therrien AS, Bastian AJ (2015) Cerebellar damage impairs internal predictions for sensory and motor function. *Current Opinion in Neurobiology* 33:127–133.

Thoroughman KA, Shadmehr R (1999) Electromyographic correlates of learning an internal model of reaching movements. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 19:8573–88.

Torres-Oviedo G, Ting LH (2007) Muscle synergies characterizing human postural responses. *Journal of neurophysiology* 98:2144–56.

Vazquez A, Statton MA, Busgang SA, Bastian AJ (2015) Split-belt walking adaptation recalibrates sensorimotor estimates of leg speed, but not position or force. *Journal of Neurophysiology* p. jn.00302.2015.

Wagner MJ, Smith MA (2008) Shared internal models for feedforward and feedback control. *The Journal of neuroscience* 28:10663–73.

Wolpe N, Ingram JN, Tsvetanov KA, Geerligs L, Kievit RA, Henson RN, Wolpert DM, Rowe JB (2016) Ageing increases reliance on sensorimotor prediction through structural and functional differences in frontostriatal circuits. *Nature Communications* 7:1–11.

Wolpert DM, Miall RC, Kawato M (1998) Internal models in the cerebellum. *Trends in Cognitive Sciences* 2:338–347.

Yokoyama H, Sato K, Ogawa T, Yamamoto SI, Nakazawa K, Kawashima N (2018) Characteristics of the gait adaptation process due to split-belt treadmill walking under a wide range of right-left speed ratios in humans. *Plos One* 13:e0194875.

Yousif N, Diedrichsen J (2012) Structural learning in feedforward and feedback control. *Journal of Neurophysiology* 108:2373–2382.

Zarahn E, Weston GD, Liang J, Mazzoni P, Krakauer JW (2008) Explaining savings for visuomotor adaptation: linear time-invariant state-space models are not sufficient. *Journal of neurophysiology* 100:2537–48.

Muscle name	Abbrev.
Tibialis anterior	TA
Peroneus longus	PER
Medial Gastrocnemius	MG
Lateral Gastrocnemius	LG
Soleus	SOL
Biceps Femoris	BF
Semitendinosus	SMT
Semimembranosus	SMB
Rectus Femoris	RF
Vastus Lateralis	VL
Vastus Medialis	VM
Sartorius	HIP
Adductor Magnus	ADM
Gluteus Medius	GLU
Tensor Fasciae Latae	TFL

Table 1. List of recorded muscles & abbreviations.

ID	Sex	Age (y.o.)	Mid speed (m/s)
C01	F	46	0.94
C02	F	51	1.02
C03	F	65	1.08
C04	F	58	0.9
C05	M	57	1.04
C06	M	52	1.05
C07	M	78	0.66
C08	F	52	1.16
C09	M	68	0.85
C10	F	62	0.98
C11	M	75	1.11
C12	M	57	0.99
C13	M	52	1.16
C14	M	64	1.25
C15	M	74	1.11
C16	M	49	1.08
Avg.	10M/6F	60 ± 9.9	1.02 ± 0.14

Table 2. Subject summary. Bottom values indicate mean $\pm$ standard deviation.

Epoch	[FHS-STO] (ms)	swing SLOW (ms)	[SHS-FTO] (ms)	swing FAST (ms)
B	168 ± 12	393 ± 59	166 ± 29	383 ± 43
eA	177 ± 46	314 ± 64	135 ± 34	458 ± 128
lA	176 ± 21	329 ± 38	161 ± 30	473 ± 114
eP	150 ± 35	358 ± 91	188 ± 32	372 ± 69
lP	187 ± 24	405 ± 42	181 ± 33	394 ± 52

Table 3. Mean time elapsed between relevant kinematic events (heel-strikes, toe-offs) during the different epochs of the experiment. Intervals are presented in order of occurrence in the gait cycle, starting at the fast leg's heel-strike (FHS). Median ($\pm$ interquartile range) values across participants.

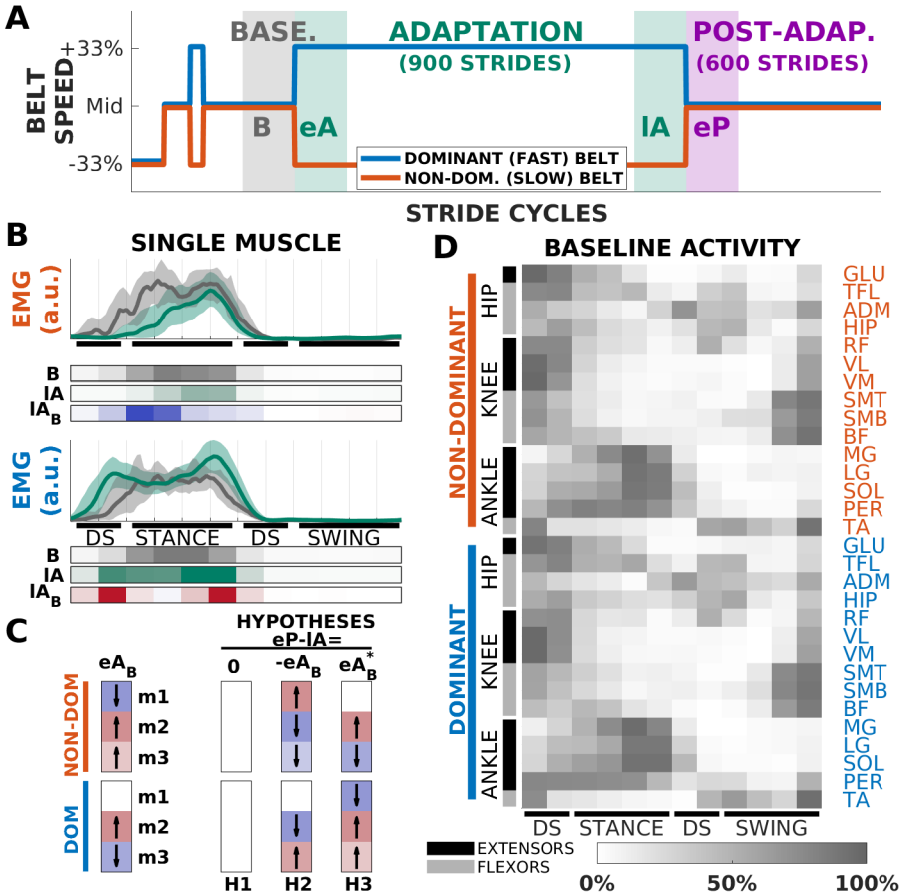

Figure 1. Summary of methods utilized in this study. (A) Schedule of belt speeds utilized. Shaded areas represent epochs that will be the focus of analysis: Baseline *B*, early Adaptation *eA*, late Adaptation *lA* and early Post-Adaptation *eP*. (B) Sample EMG traces of one muscle (LG) during Baseline (gray) and late Adaptation (green) for a representative subject (C14). Median activity across strides (lines), and the 16-84 percentile range (shaded). Colorbars below the traces represent averaged normalized values during 12 kinematically-aligned phases of the gait cycle (see Methods) for Baseline (*B*, gray), late Adaptation (*lA*, green), and the difference ($lA_B = lA - B$, red indicates increase, blue decrease). Top panels: data for non-dominant/slow leg. Bottom panels: dominant/fast leg. (C) Summary of hypotheses on feedback-mediated activity following removal of the perturbation ($eP - lA$). See Methods for a full description and rationale. (D) Summary of muscle activity during Baseline walking for all muscles, averaged as in panel B. Median across subjects. The activity of each muscle is aligned to start at ipsilateral heel-strike. Therefore, each column of muscle activity variables is synchronous for all muscles in the non-dominant (top panel) and dominant (bottom panel) legs separately, but not across legs.

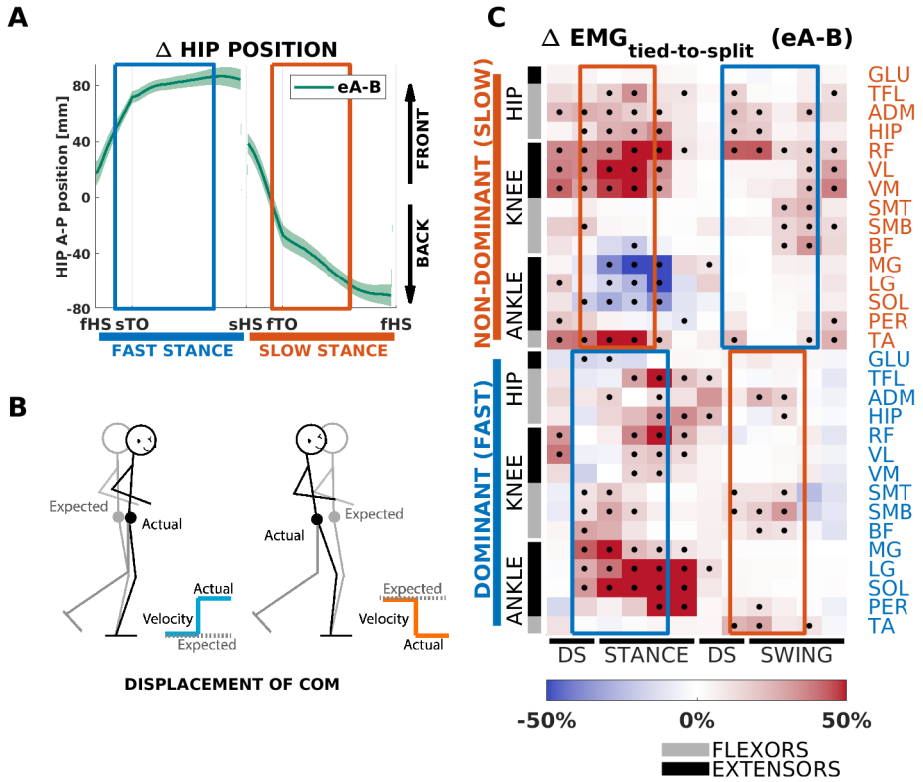

Figure 2. Muscle activity during Early Adaptation reveals coordinated feedback-mediated activity to recover balance in response to changes in environmental dynamics. (A) Relative position of the hip in the anterior-posterior direction, with respect to the (leading) stance ankle during early Adaptation, averaged across subjects. Baseline values are subtracted to illustrate the kinematic changes elicited by walking in the split-belts condition. Positive values represent the hip further ahead from the ankle. Colored rectangles indicate the interval between 100 to 400ms after heel-strike. (B) Depiction of postural differences between early Adaptation (black stick figures) and Baseline (gray) behavior. COM data is consistent with subjects 'falling' forward (with respect to prior expectation) when standing on the fast belt, and backwards on the slow leg. (C) Modulation of muscle activity during early Adaptation with respect to Baseline (median across subjects). Red colors indicate increases in activity, while blue colors indicate decrease. Dots indicate significant differences (FDR controlled, p-value threshold: $p = ...$) (see Methods). Colored rectangles indicate the same interval as in panel (A), where postural responses are expected to be present. Postural responses to 'falling' can be observed in the data. For example, TA on the slow leg is active during stance, which does not occur during normal walking. This activity is consistent with pulling the COM forward, closer to its expected position.

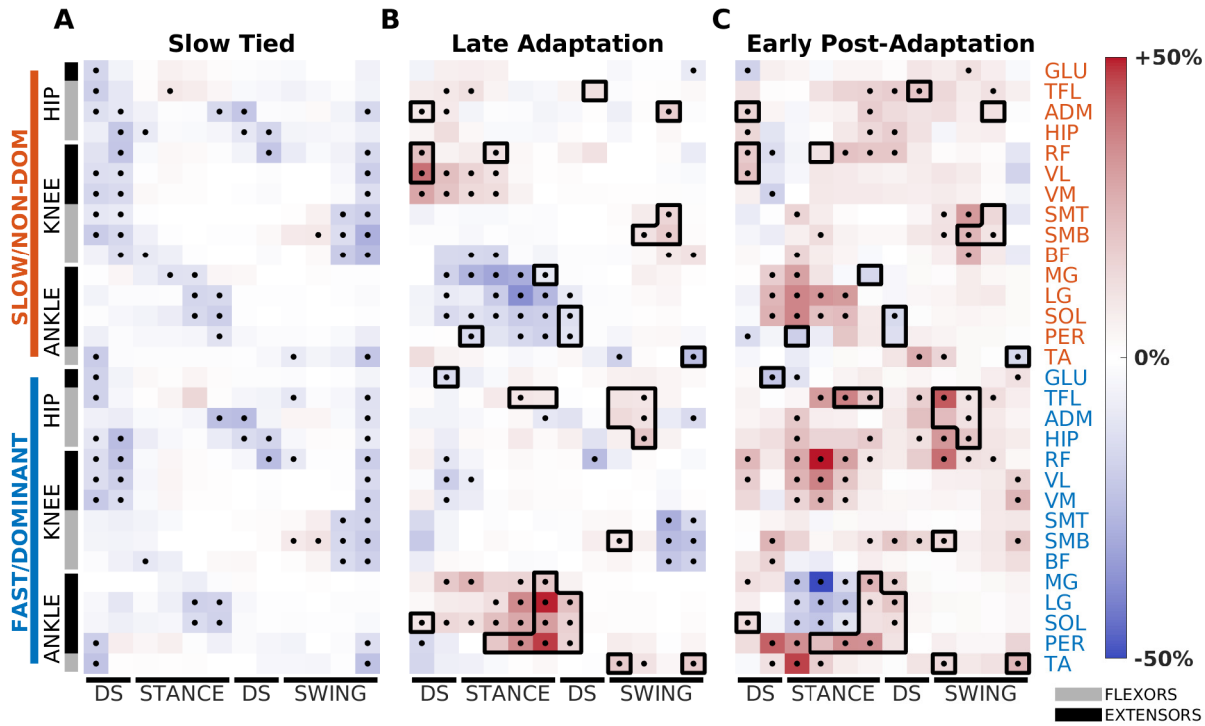

Figure 3. Steady-state muscle activity during split-belt walking and aftereffects. Panels reflect differences in muscle activity during three distinct epochs of walking, with respect to the reference (Baseline) condition. Colormap reflects effect size and dots indicate significant differences (see Methods). (A) Muscle activity modulation during slow tied-belts walking. Most muscle-phases show reduction of activity, consistent with a monotonic link between walking speed and muscle activity amplitude. (B) Muscle activity modulation during late Adaptation. Broadly, patterns of activity are anti-symmetric, with groups of muscles increasing activity in one leg and decreasing contralaterally. Those patterns do not match the expectation from simple ipsilateral speed modulation. For example, activity increases in knee muscles on the slow leg (VM, VL, RF, BF, SMT, SMB). (C) Muscle activity modulation during early Postadaptation. Few similarities are found between the steady-state activity during late Adaptation and observed aftereffects. Black lines highlight muscle-phases with the same modulation sign (i.e. increase or decrease with respect to Baseline) for both late Adaptation and early Post-Adaptation. We propose these patterns of activity (aftereffects) are generated by a superposition of feedforward- and feedback-generated activity.

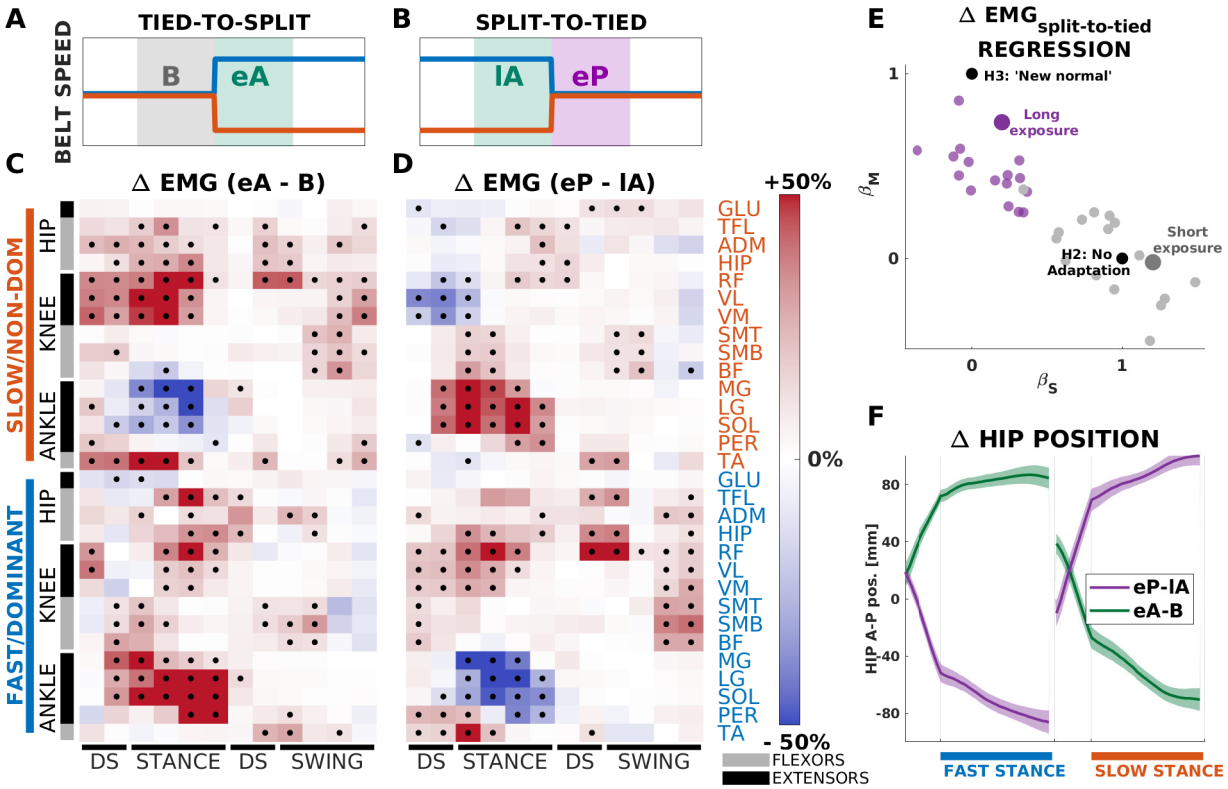

Figure 4. Comparison of changes in muscle activity following two environmental transitions. **(A)** Tied-to-split belt-speed transition. **(B)** Split-to-tied belt-speed transition. **(C)** Tied-to-split changes in EMG data. Same as Figure 2C. **(D)** Split-to-tied change in EMG data (early Post-Adaptation with respect to late Adaptation). We observe that changes in activity in the fast (slow) leg during this transition resemble those of the slow (fast) leg during the tied-to-split transition (mirroring). For example, TA is not active during stance in steady-state walking (see Figures 1D and 4A,B), but it is active following the tied-to-split transition in the slow leg, and the split-to-tied transition in the fast leg. **(E)** Quantification of the validity of H2 and H3 in split-to-tied transitions, after the short exposure (SE, gray) and long exposure (LE, magenta) to the split-belts environment. Large dots represent regressor values for mean subject data, small dots represent regressors for individual subjects. Bars indicate median and interquartile range for individual subject regressions. **(F)** Changes in hip position (with respect to previous steady-state) after the two transitions (tied-to-split in green, split-to-tied in magenta). Line represents mean across subjects, shaded area is standard error. We observe these the kinematics of these two transitions are approximately mirror images of one another, possibly causing the observed mirroring of EMG responses. For comparison, we also present early Post-Adaptation with respect to Baseline (gray).

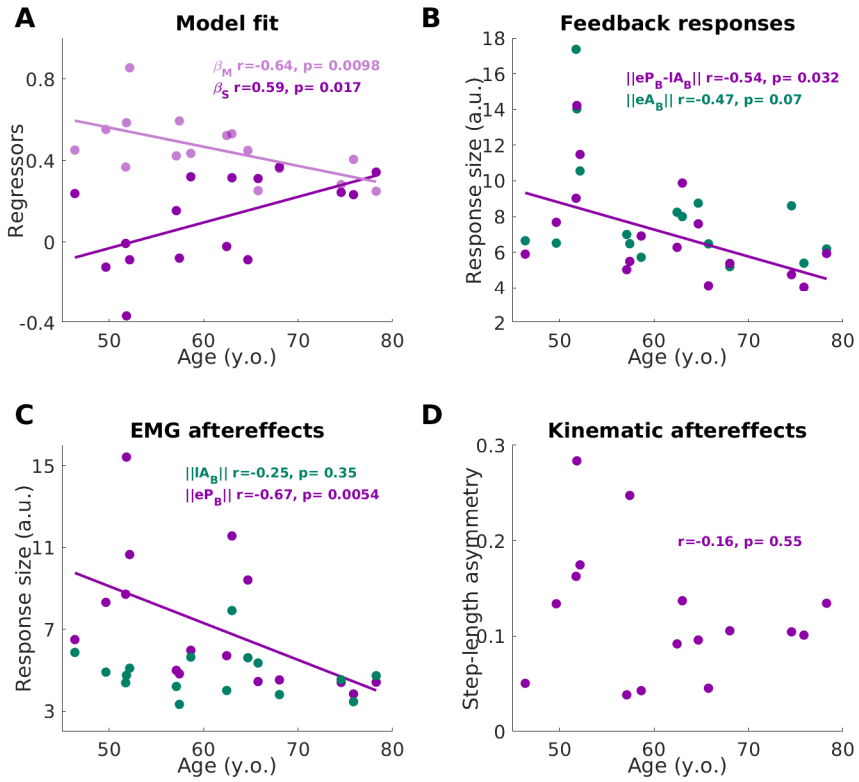

Figure 5. Correlation of age to various task performance and learning outcomes. Single dots represent values for one subject. Spearman's correlation coefficients (r) and p-values (p) are presented on the legend. When significant (p<.05) the best line fit of the dependent variable onto age is presented. **(A)** Regressors from model used to test our hypothesis on feedback responses in muscle activity (dark, β_M, light, β_S). Both regressors are significantly correlated with age. **(B)** Size of muscle activity change following transitions in environmental conditions (feedback responses). Initial feedback responses ($\|eA_B\|$)are not correlated with age, but when returning to tied-belts ($\|eP_B - lA_B\|$) there is a small age effect. **(C)** Size of muscle activity modulation during late Adaptation (steady-state) and early Post-adaptation (aftereffects). Aftereffects, but not steady-state, is correlated with age. This confirms that older subjects are able to modulate muscle activity as much as healthy subjects, but display smaller aftereffects. **(D)** Step-length asymmetry aftereffects are not correlated with age. This shows EMG-based measures of learning are more sensitive than kinematic-based ones.

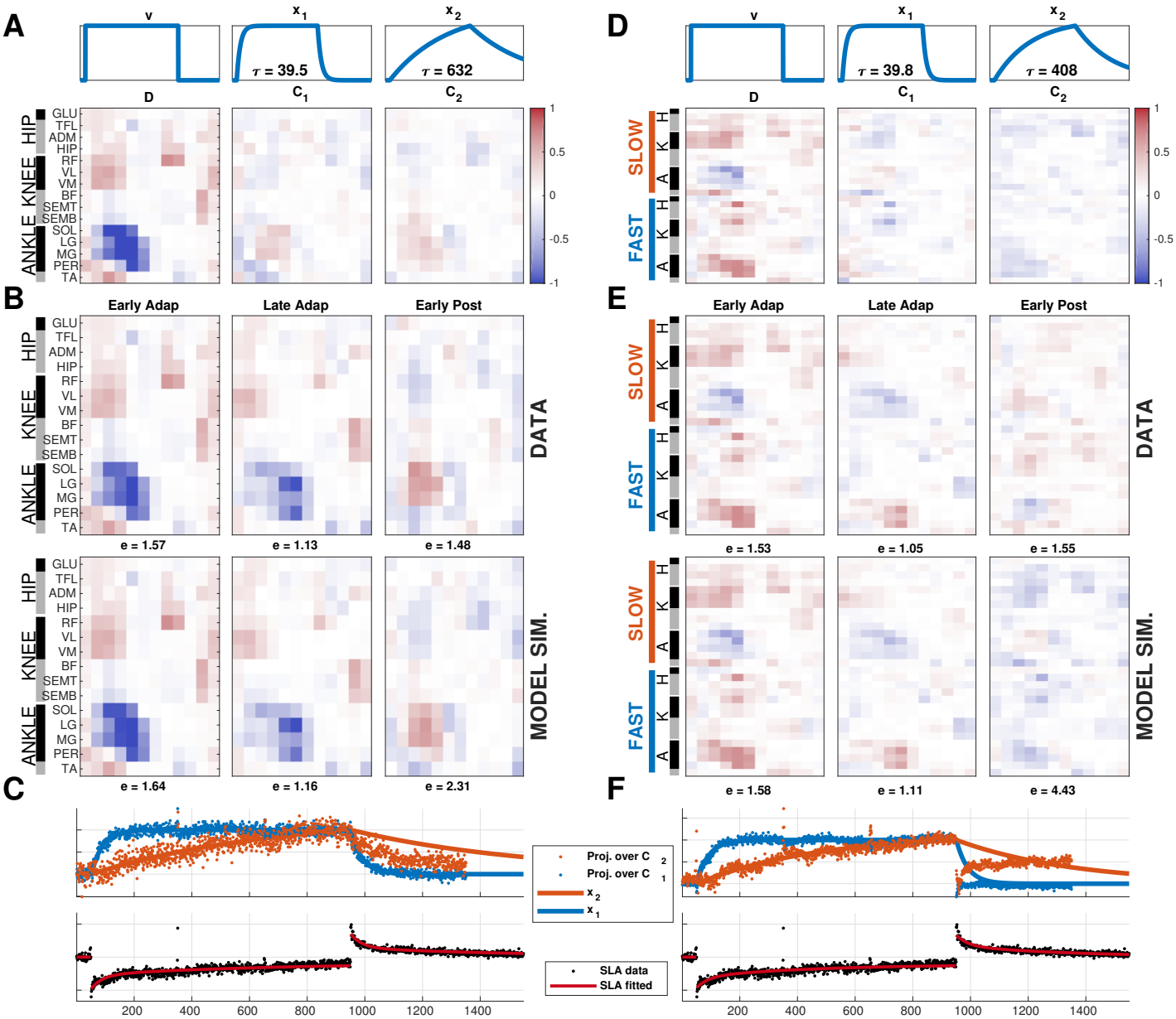

Figure 6. Two-state LTI models predict muscle activity asymmetry aftereffects but not individual leg data. **(A,B,C)** Results for the model fitted to muscle activity differences across legs. **(D,E,F)** Results for the model fitted to bilateral muscle activity. **(A,D)** Top row: temporal profiles of the signals contributing to the output. First panel corresponds to the input signal (v), and the other two to the fitted states. Bottom row: visualization of the matrix D and the columns of matrix C, which quantify the contribution of each state and the input signal onto the output (muscle activity). **(B,E)** Top row: empirical data for the three main epochs used in the analysis. Bottom row: model simulation for the same epochs. Quantification of model prediction error is displayed at the bottom of each panel (see Methods). The asymmetry model (B), but not the individual legs model (E) is able to predict aftereffects. **(C,F)** Top row: temporal evolution of the states fitted (lines) and projection of empirical data onto the subspace of each state (dots). Bottom row: step-length asymmetry (SLA) data (black dots) and